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Human Delta-type opioid receptor (OPRD1) ACTOne™ Stable Cell Line

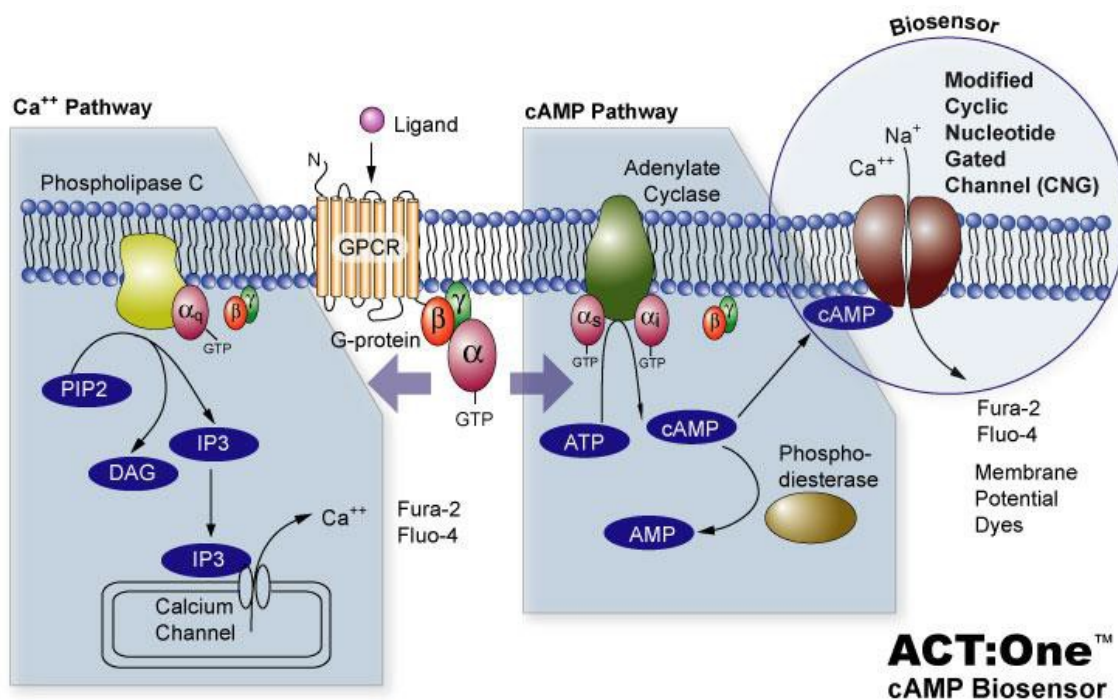
CATALOG NUMBER: CL-11-OPRD1

Introduction

OPRD1 is a G-protein coupled receptor that functions as receptor for endogenous enkephalins and for a subset of other opioids. Ligand binding causes a conformation change that triggers signaling via guanine nucleotide-binding proteins (G proteins) and modulates the activity of down-stream effectors, such as adenylate cyclase. Signaling leads to the inhibition of adenylate cyclase activity. It inhibits neurotransmitter release by reducing calcium ion currents and increasing potassium ion conductance. It plays a role in the perception of pain and in opiate-mediated analgesia. It also plays a role in developing analgesic tolerance to morphine.

Description

Human OPRD1 ACTOne™ is a HEK293-CNG cell line that expresses recombinant human HTR1A. HEK293-CNG cells express a modified CNG (Cyclic Nucleotide Gated) channel that opens in response to elevated intracellular cAMP levels and consequently result in ion flux and cell membrane depolarization which can be easily measured with fluorescent Membrane Potential Dye (Cat# CA-M145). The assay allows both end-point and kinetic measurement of intracellular cAMP changes with a FDSS, FLIPR, or a fluorescence microplate reader.



Parental Cells

HEK-293 CNG cells (originally developed by BD Biosciences by introducing CNG in HEK-293 cells) (Cat# CL-03-PC20)

Gene/Enzyme Introduced

OPRD1 (NCBI protein database NP_000902)

Applications

- cAMP dependent cell based assay for Gi-coupled human G-protein coupled receptor (OPRD1)
- cell-based high-throughput screening of human OPRD1 receptor agonists/antagonists

Mycoplasma Contamination Test

This cell line has been tested negative for *Mycoplasma sp.*.

Growth Properties

Adherent

Cell Culture Medium

- Growth medium (for OPRD1 stable cells): 90% DMEM, 10% FBS, supplemented with 250 µg/ml G418, 1 µg/ml Puromycin
- Freezing medium: 10% DMSO, 90% FBS

DATA EXAMPLE

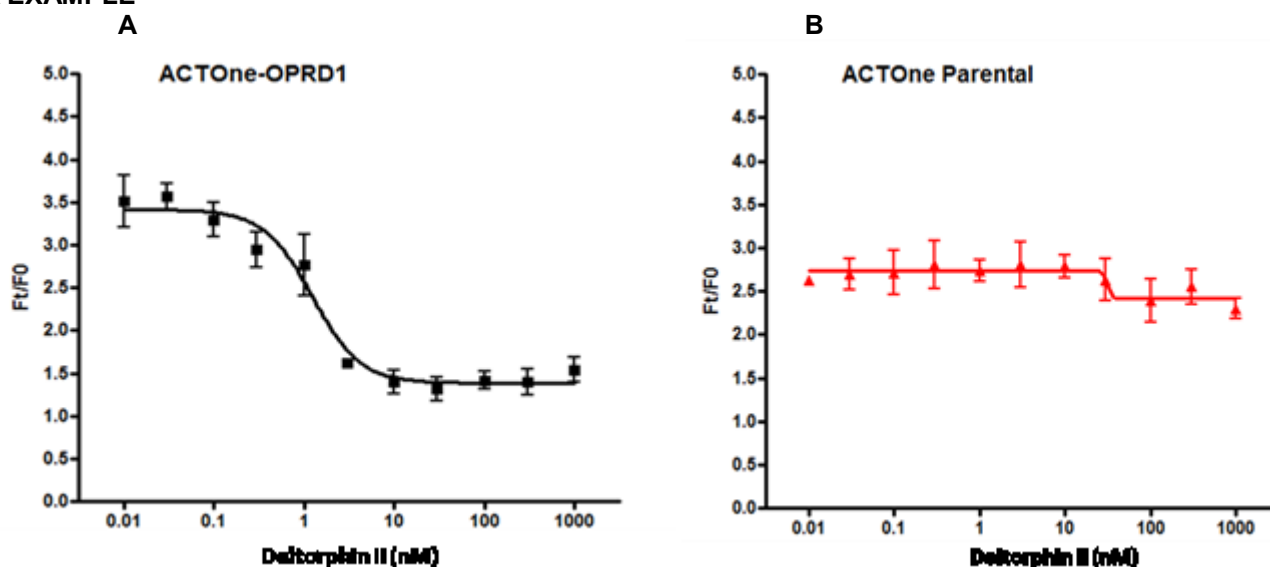


Figure 1. Response of ACTOne OPRD1 cell line & parental cell line to Deltorphan II.

ACTOne OPRD1 cells and parental cells (Cat# CL-03-PC20) were plated overnight in 20 µl culture medium on a BD Biocoat 384 well plate. The next day, cells were dye-loaded with 20 µl/well of 1X Dye-loading solution (Membrane Potential Assay Kit). After 2 hours of incubation at room temperature, two readings were obtained prior to and 40 min after the addition of DAMGO. Ratios of the two readings (F/F₀) are plotted in the figures.

- A. Dose response curve of Deltorphan II in ACTOne OPRD1 cell line. EC₅₀ = 1.2 nM in the presence of PDE inhibitor Ro20-1724 and β-adrenoceptor agonist isoproterenol.**
- B. Parental cells do not respond to Deltorphan II.**

Notice to Purchaser

1. This cell line is to be used for research purposes only. It may not be transferred to third parties, resold, modified for resale, or used to manufacture commercial products or to provide a service to third parties without written approval of eEnzyme LLC.
2. Refer to the license agreement for details on the usage restrictions.